Rejections under 35 U.S.C. §112, second paragraph

Claims 2-5 are rejected for indefiniteness, with the Examiner stating, "[I]t is unclear what is meant by the language 'different ranges of specificity' in claim 2." Applicants traverse this rejection.

As noted in the previous replies, the definition of "range of specificity" is given on page 6 of the specification. This definition reads:

> By "range of specificity" is meant the range of nucleic acid template:PCR primer ratios at which template sequences differing by at least one nucleotide may be discriminated by assaying for the presence of detectable PCR amplification product formation.

Applicants note that "specificity" in this context means the ability to amplify a target sequence preferentially over non-target sequences. The "range" of this term refers to all ratios of the amount of nucleic acid template to the amount of PCR primers that may be used to amplify a desired nucleic acid sequence preferentially over other nucleic acid sequences in a sample.

This concept can be illustrated as follows. Assume a sample contains 10 molecules of a desired nucleic acid sequence and 10 to 30,000 molecules of a PCR primer. Based on these values, the maximum and minimum ratios of the amount of target sequence to the amount of PCR primer can be calculated: (1) 10/10 = 1 and (2) 10/30,000 = 3.33×10^{-4}. These ratios form the upper and lower limits on the range of 3.33×10^{-4} to 1. Further assume that, when the desired nucleic acid sequence is amplified with the primer over this range of ratios, the desired nucleic acid sequence is the only detectable nucleic acid sequence in the product. Thus, the range of these ratios would be specific for the desired nucleic acid sequence, constituting a "range of specificity."

Because the upper and lower limits of a range of specificity for a given nucleic acid sequence and primer depend on the identity of the nucleic acid sequence and the identities of the primers, use of two different primer sets can result in "different ranges of specificity" as recited in claim 2.

In addition, the upper limit (1) of this illustrative example is 3000 times the lower limit (3.33×10^{-4}). Thus, such a range is termed a 3000-fold range of specificity.

One skilled in the art would readily understand the meaning of these terms based on the specification, and the descriptions given in this and previous replies. In view of the foregoing, the rejection of claims 2-5 for indefiniteness should be withdrawn.

Rejections under 35 U.S.C. § 102(e)

Claims 1, 6-10, and 12-20 stand rejected for anticipation by Newton. Applicants respectfully traverse this rejection.

Claims 1 and 15, the independent claims rejected, are directed to a method and a kit for determining whether a nucleic acid sequence comprises a particular allele of a polymorphic sequence. Each of these claims requires first and second pairs of primers, characterized as follows:

> (i) one of said first pair of PCR primers is (a) complementary at its 3'-terminal nucleotide to a first allele of said polymorphic sequence ... and (c) non-complementary to said nucleic acid sequence at a single nucleotide that is disposed within the five nucleotides adjacent to said 3'-terminal nucleotide ...
>
> (ii) one of said second pair of PCR primers is (a) complementary at its 3'-terminal nucleotide to said first allele of said polymorphic sequence ... and (c) non-complementary to said nucleic acid sequence at one or more

nucleotides that are disposed within the five nucleotides adjacent to said 3'-terminal nucleotide

Thus, claims 1 and 15 require two primers (one from each set) that have the same nucleotide at their 3' termini and contain at least one mismatched base. In addition, both sets of primers are capable of amplifying the same allele.

In order to anticipate a claim, a single reference must teach each and every element of the claim (M.P.E.P. § 2131). This standard has not been met in the present case.

In making the rejection, the Office relies on the teachings of Newton at column 4, lines 33-53, stating "[t]he diagnostic primer [of Newton] has the same structure as the one of said first to fourth pair of primer and one of said second pair of primer." This statement is incorrect. Newton states:

> In a second embodiment of the present invention the said sample is treated together or sequentially with either
>
> (a) a first diagnostic primer having a sequence substantially complementary to a diagnostic portion of a first nucleic acid sequence, the first diagnostic primer having a terminal nucleotide complementary to the said suspected variant nucleotide, and a second diagnostic primer having a sequence substantially complementary to a diagnostic portion of a second nucleic acid sequence, the second diagnostic primer having a terminal nucleotide complementary to the complementary suspected variant nucleotide; or
>
> (b) a first diagnostic primer having a sequence substantially complementary to a diagnostic portion of a first nucleic acid sequence, the first diagnostic primer having a terminal nucleotide complementary to the normal nucleotide which corresponds to the said suspected variant nucleotide, and a second diagnostic primer having a sequence substantially complementary to a diagnostic portion of a second nucleic acid sequence, the second diagnostic primer having a terminal nucleotide complementary to the normal nucleotide which corresponds to the said suspected variant nucleotide;
>
> the said terminal nucleotide of the first diagnostic primer and the said terminal nucleotide of the second diagnostic primer being either both at the

> 5' end or both at the 3' end of the respective primers and the first nucleic acid sequence being in the opposite sense to the second nucleic acid sequence.
>
> In this embodiment therefore, the second diagnostic primer may be considered to be an amplification primer as referred to above and hereinafter. (col. 4, ll. 31-58)

Newton's second embodiment is directed to the use of one of two possible sets of primers, one set for the variant nucleotide and one set for the normal nucleotide. This embodiment of Newton is therefore capable of detecting both alleles, i.e., normal and variant, of a nucleic acid sequence. The diagnostic primers of these two sets do not have the same terminal nucleotide because one diagnostic primer is complementary to the normal nucleotide, and one diagnostic primer is complementary to the variant nucleotide. The instant methods, however, require the use of two primers that bind to the same allele, e.g., either the normal nucleotide or the variant nucleotide, but not both. Thus, the second embodiment of Newton, relied upon by the Office, does not anticipate the instant claims.

The Office further relies on teachings of Newton that mismatched bases may be present in primers (col. 6, ll. 27-29) and on a listing of primers containing mismatched bases that are specific for an allele (col. 29, ll. 18-31). While Newton discloses a series of primers containing mismatched bases that have the same 3' terminal nucleotide, Newton only assays these primers for their ability to discriminate between a normal and variant allele to determine a single primer for use in their methods (Example 4, especially col. 33, ll. 23-34). The mere presence of primers that may be employed in the method of claim 1 cannot anticipate the method as a whole. Newton does not ever teach the use of two primers that are complementary to the same allele in a method for determining

whether a nucleic acid sequence includes a particular allele, as recited in claim 1. Thus, Newton does not anticipate claim 1, and the rejection should be withdrawn.

Regarding claim 15, the Office states that "Newton et al. also disclose constructing a kit including these primers [described above] (See column 8, lines 62 to column 9, lines 1-34)." Newton teaches that this kit may have multiple primers for (1) different diagnostic portions of a nucleic acid sequence (col. 8, ll. 45-46) and (2) both diagnostic primers from the second embodiment, described above, (col. 9, ll. 10-13). A kit containing multiple primers specific for different portions of a nucleic acid sequence clearly does not anticipate the instant claims which are directed to a kit containing two primers for the same diagnostic portion. In addition, as stated above, although the two diagnostic primers in the second embodiment of Newton are specific for the same diagnostic portion of a nucleic acid sequence, they have <u>different terminal nucleotides</u>. In contrast, two primers in the kit of claim 15 have the <u>same terminal nucleotide</u>. Since the primers in the kit of Newton are not the same as the primers in the instant claims, Newton does not anticipate claim 15, and this portion of the § 102 rejection should also be withdrawn.

CONCLUSION

Applicants submit that the claims are in condition for allowance, and such action is requested. If there are any charges, or any credits, please apply them to Deposit Account No. 03-2095.

Respectfully submitted,

Date: 9/16/03

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